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## Table of Contents

I	age
Summary	1
Background	3
Method	3
Comparative assays of lunar samples and initial confirmation of our results	4
Integrated results	5
Amino acids as precursors	7
Ratio of amino acids/carbon	7
Analyses relative to jet exhaust	8
Glassy microparticles	11
Significance of assays of amino acids in lunar fines	12
Unanswered questions for future research	16
Bibliography for this report	18
Publications from this program, NGR 10-007-008	20
Long abstracts from lunar program	23
Popular presentations	24

# Final Technical Report NASA Grant Number NGR 10-007-088

#### SUMMARY

Analyses of twelve collections from the six Apollo missions have shown a consistent pattern of amino acids obtainable from the lunar dust. These amino acids are six of the twenty always found in the total proteins of contemporary terrestrial organisms. The amounts found represent 0.005% to 0.10% of the carbon in the samples. The results have been confirmed by two other lunar analysis teams (Nagy's and Ponnamperuma's). The amino acids obtained have been shown not to be due to contamination from humans, from jet exhaust, nor from other nonlunar sources.

The evidence, in association with data from other investigations, suggests that the compounds yielding amino acids have been implanted into the surface of the Moon from the solar wind. It thus appears that some of the earliest steps in chemical processes that led to life on Earth have occurred on the Moon. Since it has become possible, on the basis of laboratory experiments, to explain the origin of primordial life on Earth, it is now possible to state that life arose on Earth because of the presence of sufficient water, which has been crucially lacking on the Moon. For primitive life to have arisen on the Moon, however, the mere addition of water would have not been adequate.

The kinds and amounts of amino acids, relative to carbon, found on the Moon have been closely similar to those found in several meteorites

analyzed by the special sampling and analysis that our group first introduced for Apollo 11 fines.

Since the main protein-related amino acids found in such meteorites resemble those found in the lunar samples, we can conclude that there is a common cosmochemical pattern in two extraterrestrial sources, Moon and meteorites, of different cosmophysical histories. This provides the first solid evidence of a common course of cosmochemical reactions for carbon and is especially significant in identifying a somewhat, or highly, common cosmochemical matrix for the unity of biochemistry. The unity of biochemistry has been firmly established in the past few decades by biochemical analyses. We can say, in a grand overview and on the basis of evidence, that we live in an orderly Universe.

The most accurate advance estimate of the amount of amino acids (precursors) to be found in lunar dust was evidently made by our group.

We had, under NASA funding, studied fresh terrestrial lava in the 1960s and had found amounts of amino acids per weight of sample that proved to be approximately ten to a hundred times as large as those subsequently found in the first lunar samples. Both a realistic anticipation of amount of total lunar amino acids and a developed method of sample preparation was thus transferred from studies of terrestrial lava to those of lunar dust.

Much as the technology for lunar dust was subsequently applied to meteorites, we can anticipate that we are better prepared to transfer lunar data and methods to Mars, as well as interpretations of data therefrom. We have good reason to anticipate that the values found on Mars will be bracketed by those from the Moon and from the Earth.

## Background

The expectation for organic compounds on the Moon was characterized originally (Ponnamperuma, 1972) as "a bonanza of organic molecules."

Following the first reports on Apollo 11 lunar fines, premature publicity stated that there was nothing on the Moon. This swing can be viewed as the well-known pendulum effect in science.

These laboratories did not expect a bonanza of organic compounds. Our expectations were shaped in two ways. Firstly, the laboratory experiments which were invoked for the "bonanza" employed gas-tight flasks. We felt that this kind of condition was too far removed from what could be expected on the surface of the Moon. Secondly, our own analyses, of many years, of basalt from various volcanic regions gave us a sense of what might be expected and how much, and especially provided practical information on suitable methodology to employ. The amounts of amino acids obtainable from cooled terrestrial lava, which had once been at 1000°C, in no case exceeded three ppm. Moreover, the pictures from the Surveyor program suggested a similarity to many of the volcanic sites on Earth. This exercise with terrestrial lava also provided a practical sense of the way in which contamination might appear, and how to avoid it. The proportions found in lunar samples were 1/10 to 1/20 as much as found in most terrestrial lava samples.

#### Method

The essence of a practical method of estimation of small quantities of amino acids, which were already recognizable as being present as

precursors, was to extract the sample with hot water and then hydrolyze the aqueous extract. This maneuver and procedure permitted avoiding both the decomposing action of a great excess of minerals and the physical loss which could occur through the formation of large excesses of salt resulting from direct hydrolysis of samples.

Other requirements, it subsequently developed, were that the extracts had to be done with hot water and that the hydrolyzates had to be estimated by a sensitive assay instrument.

## Comparative assays of lunar samples, and

## initial confirmation of our results

Until Apollo 14, substantial disagreement existed between the various groups analyzing lunar fines for amino acids or their precursors. The Nagy group and our team were in substantial agreement on free amino acids and their nature in the Apollo 11 fines, and were in substantial agreement on the absence of free amino acids in the Apollo 12 samples.

Following unsatisfactory comparisons between laboratories, our team suggested a side-by-side comparison in one laboratory. In order to avoid the problem of contamination, presumed by some to have been introduced at the LRL, both Dr. Harold Urey and the present PI requested a special sample from Apollo 14, which was opened in the cleanroom at Berkeley. At the present time, following the studies, we can say that the LRL was operated in a totally clean manner, insofar as the single criterion of contamination by amino acids is concerned.

A significant confrontation in the analysis of this SESC sample occurred in October 1971 at the Ames Research Center where the analysts

in our group ran assays side-by-side with the analysts in the Ponnamperuma group, who had disagreed with our findings. This comparison was a test of the two instrumental methods of assay, the GLC on one hand, and the IEC according to Hare, on the other. It was not a suitable test of two methods of sample preparation, since at the site of the Ames Research Center all analysts present agreed that Dr. Harada of our team should prepare clean water or that they use that which he had prepared. Dr. Harada also prepared the samples for analysis. These samples were then analyzed by the GLC method by Gehrke and his associates and by the IEC method by Hare. The results were in substantial agreement, as recorded in the October 1972 issue of Space Life Sciences (Fox et al., 1972; Gehrke et al., 1972).

However, the results showed that, at these levels, the GLC method revealed more than twice as many peaks as expected from the six amino acids found. Some of these unexplained peaks were considerably larger than the peaks found in confirmation of the amino acids earlier designated by IEC. Accordingly, the IEC method could be used for primary identification, but the GLC could be used only for confirmation.

#### Integrated results

The analytical results from eleven collections from six Apollo missions are presented in Table I. A twelfth result, representing a trench sample from Apollo 12, no. 12001, is not included because it is known to have been contaminated by the entrance of unclean air into a flask in the analytical laboratory. That sample was distinct in having a few percent of the total present as tyrosine and phenylalanine. The dominant amino acids in 12001 gave essentially the same profile as the samples of all other collections (Table I).

. 1

 $\frac{a}{1}$ 

			o me tulo					Total
Sample	Glycine	Alanine	Acid	Acid	Serine	Threonine	Others	6/bu
10086	20	25	σ	Ŋ	σ	8	ပ	45
12033	49	16	27	~	7	7	Ŋ	19
14003	62	20	12	7	4	п	0	19
14163	47	26	20	2	9	4	0	30
14240	63	15	ø	11	4	0	0	ca. 5
14298	57	7	13	7	01	8	4	37
15012 <sup>2/</sup>	61	9	16	9	9	7	m	7
15013 <u>b</u> /	73	œ	м	7	7	8	4	12
66041	99	ø	19	2	œ	1	Ŋ	12
70011 <u>5</u> /	83	12	0	м	4	0	н	30
72501 <u>c/</u>	70	n	7	7	m	<b>~</b>	0	10

All calculations exclude ammonia and basic amino acids (the latter are essentially absent).

b/ Proximal sample

<u>c/</u> Distal sample

Our results on amino acid precursors have been consistent throughout the entire series, beginning with Apollo 11. On Apollo 11 fines, our results on free amino acids are in qualitative agreement with those of Nagy et al. (1970). On Apollo 12 fines, both Nagy et al. and we found no free amino acids, but again we found the amino acid precursors. On the Apollo 14 SESC sample, the Nagy group (Modzeleski et al., 1973) obtained essentially the same results, as had we and Gehrke et al. (1972).

On Apollo 17, Gehrke et al. (1974) obtained the same results as we.

## Amino acids as precursors

The amino acids obtained from the various analyses appear primarily as the consequence of hydrolysis of hot aqueous extracts. This signifies that the amino acids are in some chemical precursor (probably also evolutionary precursor) form, and are released as free amino acids upon hydrolysis. The amino acid-like compounds of fresh terrestrial lava and the primary products of chemical synthesis in the laboratory appear also to be precursors. Only in those cases in which hot water is present and retained within a flask for long periods, as in the Miller synthesis (1953), are free amino acids found. The free amino acids appear to be a consequence of hydrolysis. We believe that the small amounts of free amino acids observed in some lunar samples are also the result of extraction by hot water in the presence of catalytic lunar minerals.

The fact that amino acids are present as precursors has several significances, to be discussed later in this report.

#### Ratio of amino acids/carbon

The proportions of total amino acids in the lunar samples was small, less than 50 ppb, or less than 50 ng of total amino acids per g of sample.

The question can therefore be raised of the significance of such small amounts; this question has been raised especially in comparison with the content of proteinous amino acids in meteorites.

The question is properly phrased in terms of carbon content, since amino acids and amino acid precursors are compounds of carbon. When the comparison is made on the basis of carbon, the amounts found are comparable from the two sources; all analytical values are covered essentially by one order of magnitude, a narrow range for this kind of analysis. In other words, even though hundreds of times as much amino acids are found in 1 g of the Murchison and Murray meteorites as in lunar fines, the proportion of carbon is also hundreds of times as great. The amounts in both sources are therefore probably equally significant. Materials from each source have also similar profiles of proteinous amino acids. This comparison is especially defensible since the most advanced method of analysis used on these meteorites and on Allende is the same as introduced earlier on Apollo 11 fines. The validity of the comparison on the basis of amino acids/carbon is recognized in the Summary of Conference on Interactions of the Interplanetary Plasma with the Modern and Ancient Moon, p. 24. The results from lunar dust add credence to those from meteorites, the latter having been in doubt, inasmuch as they were recovered after passage through a contaminated terrestrial atmosphere and after resting on contaminated soil.

#### Analyses relative to jet exhaust

Samples have been collected by astronauts from beneath the descent engine (15013 and 70011) and from maximal distances on two missions (15012 and 72501), by our initial request. The distal cample from Apollo 15 was

taken at 4 km from the LM. The distal Apollo 17 sample was taken from a distance of 6-1/2 km. Since the Apollo 17 values are close to those of the Apollo 15 figures, the results are essentially confirmed. The calculations to be presented are from the Apollo 17 results.

The total distal Apollo 17 sample, 72501, was 687 g, collected to a depth of 4 cm. Since the soil has a density of approximately 1.7, 687 g would have been collected from an area of

$$\frac{687}{1.7 \times 4}$$
 cm<sup>2</sup> = 100 cm<sup>2</sup>

Since each g of soil contains 10 ng of amino acid precursor as amino acids, 100 cm<sup>2</sup> of the area 6-1/2 km in radius contained 770 x 10 ng =  $7 \times 10^3$  ng.

By rounding off 6-1/2 to 6 and  $\, \Pi \,$  to 3, we find for the total area of the circle

$$6000^2 \times 3 \text{ m}^2$$
  
= 3.6 x 3 x 10<sup>7</sup> m<sup>2</sup>  
= 10<sup>8</sup> m<sup>2</sup> = 10<sup>12</sup> cm<sup>2</sup>

The disc therefore contains at least  $\frac{10^{12}}{10^2} \times 7 \times 10^3 = 7 \times 10^{13}$  ng =  $7 \times 10^4$  g. These calculations are based on the assumption that the concentration of precursor is no greater closer to the descent engine than at the periphery of the circle; this is of course a conservative assumption.

Table XVII of the paper by Flory et al. (1972), describing potential organic contamination from jet exhaust from a model rocket engine indicates that the HCN produced is less than 5 parts per 1000 parts of the exhaust, i.e.  $5 \times 10^{-3}$ . The overall results from the model rocket were in close agreement with those obtained on the Moon itself, by Freeman et al. (1973).

Since experiments in the conversion of HCN to  $\alpha$ -amino acids show a rate of  $10^{-4}$ , the proportion of amino acids derivable from the exhaust is approximately  $5 \times 10^{-7}$ , that is:

fuel 
$$\rightarrow$$
 HCN = 5 x 10<sup>-3</sup>; HCN  $\rightarrow$  amino acids = 10<sup>-4</sup>  
5 x 10<sup>-3</sup> x 10<sup>-4</sup> = 5 x 10<sup>-7</sup>.

The figure may be larger if other compounds containing C, H, and N prove to be convertible to amino acids by hydrolysis, but those reported suggest that it can not be as much as by an order of magnitude, if any at all.

The mass of fuel used in descent was  $3 \times 10^6$  (Duke, 1974). If we assume that all of this was somehow captured for conversion to amino acid precursor on the surface of the Moon, we would expect  $3 \times 10^6 \times 5 \times 10^{-7} = 1.5$  g. The analyses and calculations indicate, however,

$$\frac{7 \times 10^4 \text{ g}}{1.5 \text{ g}} = 2.5 \times 10^4$$

too much precursor in the disc to be accounted for by conversion products of jet fuel. The amount found is thus an insignificant proportion of the fuel used.

It seems obvious that the actual conversion of fuel to exhaust trapped on the lunar soil was very small, since the predominant fraction of the fuel oxidation products must have been lost to the high vacuum of outer space during the 12-minute descent. The assumption of no such loss is, accordingly, a very conservative one.

By ignoring the exact value of the step of fuel + soil-trapped exhaust products, which is difficult to estimate, one finds that the actual conversion of exhaust to amino acids is less than that needed to explain the amount of precursor found-by approximately four to five orders of magnitude.

We conclude that chemical conversion of jet fuel to amino acids can not have contributed significantly to the amino acid precursors found in the surface. Since, as indicated earlier (Harada et al., 1971; Modzeleski et al., 1973), the amino acids obtained are much unlike that from human contamination, the amino acids obtained do not result from human contamination nor the jet exhaust. The amino acid precursors are thus indigenous to the Moon's surface (although probably derived from nonlunar and nonterrestrial sources).

## Glassy microparticles

The glassy microparticles were fractionated by rolling the rounded ones down an inclined piece of paper several times. They were then studied statistically.

The glassy particles are perhaps formed by solar flares on the lunar surface; they differ from particles generated by terrestrial volcanoes, by meteoritic impacts, or by condensation of parent bodies of meteorites.

The lunar particles were studied by transmission and scanning electron microscopy.

## Significances of assays of amino acids in lunar fines

All chemical analyses of planetary surfaces, as obtained by remote control or by analysis of returned samples, are part of the test of the theory that has been built up for the origin of life from laboratory studies. The Apollo program proved to be unexpectedly informative in providing new data and new perspectives for the concepts that have existed.

An outstanding new view which has emerged is that of the importance of being cautious in the conceptual transference of data obtained from gastight flasks to planetary surfaces. Although a number of other factors were involved, the main basis for the prior assumption of a "bonanza" of organic molecules on the surface of the Moon was experiments performed in the early 50s within which the walls of a closed flask confined gases under pressure. Within the flasks, a bonanza was indeed obtained; on the surface of the Moon no bonanza was found.

The lunar analyses also emphasize the importance of study of compounds directly convertible to amino acids by simple hydrolysis. Here too, results from experiments performed in flasks were confusing. They were misleading for the reason that they retained within them water that would otherwise have distilled from the site of reaction. In reactions that were allowed to continue for one or two weeks, with water at the boiling point, the amino acid precursors that must have been formed in the reaction flask had much opportunity to be hydrolyzed to free amino acids. The fact that free amino acids are sometimes found as a result of laboratory investigations should not therefore be construed to signify that free amino acids are formed in the experiment. The accurate picture is, rather, that compounds

of an incompletely identified type are formed and that these are rather easily hydrolyzed to amino acids.

This finding highlights another meaning. This significance is the one that amino acids in general in the terrestrial, extraterrestrial, and organismic realms are available rather predominantly as amino acid precursors. In organisms, however, the amino acids are found predominantly as conversion products from amino acids, proteins; the proportion of free amino acids in organisms is exceedingly small. The benefit of amino acid precursors to an evolutionary stream stems from the fact that free amino acids would, on the basis of structural organic chemistry, be quite unstable in an environment subjected to solar radiation. This would not be true, necessarily, for suitable precursors such as certain nitriles. Therefore, somewhat as in organisms, the amino acids are protected by being something other than free amino acids. They are, more accurately, in existence as precursors which are capable of being transformed readily to free amino acids, which are in turn transformed promptly to more stable compounds. In the evolutionary context, the evolutionary sequence can be seen to have been protected by this state of the amino acids. A realization of the importance of the amino acid precursor can thus be attributed to the Apollo program, and also to confirmation by subsequent studies with meteorites (which however contained some water, evidently, in their preterrestrial state).

Another significance of the relationship of what is on the Moon to water is the comparison between molecular evolution on the Moon and on the Earth. On the Earth there has been abundant evolution and proliferation of organisms. On the Moon there are found no organisms nor traces of any

earlier organisms. This can be understood on the basis that the Moon has had at no time any significant amount of water.

Another significance of the contribution from the Apollo program is that, in the various ways already indicated, it clarifies the first steps involved in molecular evolution toward the origin of life. The steps involving polymerization of amino acids or hydrolyzable amino acid precursors to a model for prebiotic protein and thence to protocells have been placed on a very firm basis by identification of the properties found and by the geophysical relevance of the reactions. The same has not been true for the availability of amino acids for the beginning of such an evolutionary sequence. The Apollo program has led the way to a more realistic view of the availability and forms of amino acids in the earliest steps. The principal ignorance that remains concerns those steps in between the kinds of organic compounds that are found by astrophysical studies to be present in abundance in the Galaxy, and the free amino acids used in laboratory experiments for conversion to protoinformational molecules and protocells.

Another "first" that comes out of the Apollo program is that this was the first instance in which amino acids (Apollo 11) and amino acid precursors were found in (clean) extraterrestrial samples brought to Earth.

Finally, the perhaps most significant relationship which has been placed on a solid basis by the Apollo 11 analyses and the methods developed for Apollo 11 is the indication of a considerable, or high, degree of unity in the carbon cosmochemistry in the Solar System. The similar quantities and species of amino acids obtainable both from lunar fines and meteorites

strongly indicate, despite probably very different physical histories of the two extraterrestrial sources, such a unity. This is of particular interest at this stage in science because the hypothesis of a unity of biochemistry is now on a very firm basis (Florkin and Mason, 1960). The new results indicate that the unity of biochemistry had its roots in the unity of cosmochemistry. It thus appears that we live in a very orderly Universe, and this order is observable and documentable especially at the molecular level at all stages of evolution.

## Unanswered questions for future research

The exact chemical nature of the precursors of the amino acids has yet to be established. Comparison has been made with (a) hydrolyzates of polymers of HCN, and (b) hydrolyzates of reaction products of formaldehyde and ammonia. Both chemical sources (a) and (b) represent likely extrater-restrial chemical matrices. The amino acid profile more closely resembles that from (b). Further investigation of such comparisons, indirect identification of the actual precursors and, finally, direct identification are called for. Indirect identifications are suggested first, since experiments for direct identification will probably require prohibitive quantities of lunar fines.

Observations in three laboratories indicate that the amount of amino acid precursor increases markedly when a water-extracted lunar sample is stored for months in a desiccator. This seems to occur only in the case of those samples that have been extracted at least once by water. The derivative suggestion is that the analysts are dealing with precursors of precursors. Solution of the problem posed may help to identify the preamino acids.

Experiments in the production of amino acids and amino acid precursors in lunar fines, or in artificial mineral matrices corresponding to lunar inorganic analyses, are indicated.

Simulation of solar wind implantations should be tested. This could be done with terrestrial basalt, and finally tested on lunar samples.

Since the amino acids obtained from lunar samples and from terrestrial lara are similar (Fox, 1973), further comparative studies may aid in

identification of the lunar precursors. The data should also contribute to the growing picture of comparative cosmochemistry.

With the profile firmly established for lunar fines, samples of various lunar rocks should be analyzed.

A more thorough quantitative evaluation of the conversion of jet exhaust and the types of products related to amino acids might be informative.

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## Popular Presentations

Presentations to lecture audiences, journaise, W and radio audiences have been numerous.